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PHARMACOLOGICAL PROPERTIES OF METAL (SILVER, COPPER, AND IRON) NANOPARTICLES

The paper contains summarized results of studies on the pharmacological and specific properties of nanometals (silver, iron, and copper). The acute toxicity of metal nanoparticles depends on their nature, way of administration, and sex of laboratory animals. The nanoparticle effect on the heart activity, the hemodynamic status, and the osmotic resistance of erythrocyte membranes depend on the dose administered.

Keywords: nanoparticles, nanosilver, nanocopper, nanoiron, and properties.

Both domestic and foreign scientists are conducting intensive researches of physical, chemical, biological, biochemical, and pharmacological mechanisms of action of nanoscale materials. This will facilitate the development of new nano-medications for prevention, diagnosis, and treatment of various diseases and the creation of modern cost-effective and environment friendly nanotechnologies for the production of such nanostructures [1, 2].

Today, the researchers are intensively studying new phenomena typical for the nanomaterials such as an huge magnetic resistance and dimensional quantization effects; the designers and engineers are creating new modern equipment to study the nanoparticles having a size of 0.1–100 nm (electron microscope, phase-contrast microscope, scanning tunneling microscope, etc.).

Special attention should be paid to studying the toxicity of organic and inorganic nanomaterials. The toxicity of nanomaterials depends on method of their production, size, structure, physical nature, and on biological structures affected. The scientists and researchers face an important scientific and social challenge, to conduct a thorough study of possible toxic effects of nanostructures on the living cells and on the environment, as well as to offer effective methods for safe handling of such materials and antidotes for the prevention or mitigation of their adverse effects [3].

The nanoparticles of metals (silver, copper, and iron) may be used as substances for the creation of original medications, highly sensitive diagnostic tools, and medicines for the treatment and prevention of many infectious and non-infectious diseases [4].

Among the metal nanostructures with strong biocidal properties, the most well-studied are nanoparticles of silver and copper, which are actively used in drug design and creation of medical products, synthetic fibers, packaging materials with antimicrobial effect, and means of water disinfection [5, 6].

The relevance of this matter is greatly enhanced due to the fact that, in recent years, the resistance of microorganisms to antibiotics has increased materially. Therefore, the development of antimicrobial medications based on silver and copper nanoparticles is a very promising direction.

It is known that nanosilver is a promising antimicrobial, anti-inflammatory, and immune-modu-

lating agent. Nanocopper is also a powerful antimicrobial agent that can be used for creating effective drugs for the treatment of infectious diseases [7].

Iron deficiency anemia (IDA) is the most common kind of anemia that develops in the case of absolute or relative deficiency of iron necessary to maintain normal hematopoiesis. The main causes of iron deficiency are insufficient intake of iron from food, inadequate utilization of iron in the case of chronic inflammatory diseases, malabsorption or excessive loss of this trace element. IDA is a severe medical and social threat for increasing morbidity and mortality among the population. The medicines based on nanoiron are likely to be used in the treatment of iron deficiency [8—11].

The Department of Pharmacology of the O.O. Bogomolets National Medical University, in cooperation with the institutions of the NAS of Ukraine, the AMS of Ukraine, as well as with higher medical establishments of Ukraine, has studied specific pharmacological properties of metal (silver, copper, and iron) nanoparticles to create effective antimicrobial drugs and to apply them to clinical practice.

The Department of Pharmacology collaborates with institutions of the NAS of Ukraine (the E.O. Paton Electric Welding Institute, the F.D. Ovcharenko Institute for Biocolloidal Chemistry, and the O.O. Chuiko Institute of Surface Chemistry), the NAMS of Ukraine (the Institute of Urology, the Institute of Labor Medicine, and the L.V. Gromashevskyi Institute of Epidemiology and Infectious Diseases), the Vinnitsa National Medical University, the Zaporizhia State Medical University, the Danylo of Galicia Lviv National Medical University, the Odesa National Medical University, the Chernivtsi State Medical University, the Dnepropetrovsk Medical Academy, the Ukrainian Medical Stomatological Academy, and the P.L. Shupyk National Medical Academy of Postgraduate Education. This collaboration is aimed at developing technologies for obtaining metal nanoparticles, studying their pharmacological properties, and creating medical modifications of nanosilver, nanocopper, and nanoiron.

Toxicity is a very important factor for the introduction of new drugs, so the Department has studied acute and sub-acute toxicity of nanosilver, nanocopper, and nanoiron.

As one can see from Table, the toxicity of metals depends on gender and way of administration.

The next stage of research is to study the influence of metallic nanoparticles administered intravenously on the heart activity and hemodynamic status of rabbits. The effect of silver, copper, and iron nanoparticles have been established to depend on dosage. The conventional therapeutic doses have no adverse effect on blood pressure, heart rate, cardiac minute volume, stroke volume, heart discharge, cardiac index, systolic index, total peripheral resistance index, left ventricle index, and left ventricular stroke volume index. As the dosage approach the toxic value, the above indices deviate from the norm [12].

An important part of preclinical drug research is to establish the effect of tested substance on the blood system, including on the ability of red blood cells to preserve normal structural and func-

Table 1

Acute Toxicity of Metal Nanoparticles for Intravenous and Intragastric Administration by CD-1 Mice

Nanometal	LD50 for intravenous administration, mg/kg		LD50 for intragastric administrations, mg/kg	
	Male	Female	Male	Female
Nanosilver, size 30 nm	83.2 ± 10.9	99.9 ± 11.7	34.5 ± 3.8 (intragastric)	22.1 ± 2.3 (intragastric)
Nanocopper, size 20 nm	540 ± 25	443.7 ± 53.5	2200 ± 150	1839.1 ± 210.2
Nanoiron, size 40 nm	231.4 ± 8.1	207.5 ± 10.6	>5000	>5000

tional characteristics. For this purpose, the effect of tested substance on osmotic the resistance of erythrocyte membranes (an indicator of the erythrocyte protection from hemolysis in hypotonic electrolyte solutions) has been studied. This research can establish possible adverse effects of the tested substance on the integrity and chemical structure of erythrocyte membranes.

The effect of copper nanoparticles on osmotic resistance of erythrocyte membranes of laboratory rodents has been established to depend on dosage. As their concentration exceeds 3.75 μg/ml, the hemolysis of erythrocytes in NaCl hypotonic solution increases. The effect of ionic copper ($CuSO_4$) on the osmotic resistance of erythrocyte membranes is statistically significant at a concentration of 1.88 μg/ml. This means that the copper nanoparticles have lower toxicity as compared with the ionic copper (see Table 2).

The same pattern has been recorded for the effect of silver nanoparticles on osmotic resistance of erythrocyte membranes of laboratory rodents (rats).

In terms of preclinical studies, the effect of potential drugs on blood biochemical parameters is a very important aspect. Therefore, the effect of metal nanoparticles on blood biochemical parameters has been studied in rats.

For the mice administering silver nanoparticles at doses of 1.6 mg/kg, 8 mg/kg, and 16 mg/kg, the activity of alanine aminotransferase and aspartate aminotransferase has been established to depend on dosage. As the dose ups, the activity of alanine aminotransferase increases, while that of aspartate aminotransferase decreases. Silver nanoparticles at a dose of 16 mg/kg administered intravenously significantly decrease bilirubin and creatinine concentration. For the lower doses, no statistical differences as compared with the reference group has been reported.

The iron nanoparticles have a prominent anti-anemic effect and can be used for the treatment of iron deficiency anemia. They are more effective as compared with the effect of comparator (iron (III)-hydroxide polymaltose complex). It should be noted that iron nanoparticles have anti-anemic effect both at conventional therapeutic dose and at 1/10 thereof.

The preventive use of silica nanocomposite with a concentration of silver nanoparticles of 200 μg/ml prevents creation of *P. aeruginosa* biofilms. Being applied to the existing biofilms at a concentration of 500 μg/ml it causes the film degradation without recovery of initial structures. The effect on biofilms can be associated with pronounced antimicrobial effect of silver nanoparticles.

The *in vitro* studies of antimicrobial effect of silver nanoparticles in relation to pathogenic test strains have showed their prominent effect on the strains of *Staphylococcus aureus MRSA ATCC* 43300, *Pseudomonas aeruginosa ATCC* 27853, *Escherichia coli ATCC* 2592, *Shigella sonnei*, and *Salmone-*

Table 2

Effect of Copper Nanoparticles on Osmotic Resistance of Laboratory Rodent Erythrocyte Membranes *in vitro*

Sample	Erythrocyte hemolysis (%) in NaCl solutions of various concentration				
	0.5% NaCl	0.45% NaCl	0.4% NaCl	0.35% NaCl	0.1% NaCl
Reference (n = 10)	0.55 ± 0.07	0.44 ± 0.06	10.84 ± 1.25	65.15 ± 6.88	100 ± 0
CNP 1 (n = 10), ρ(Cu) = 1.88 μg/ml	0.39 ± 0.06*	1.42 ± 0.19*	13.71 ± 1.62	71.67 ± 7.88	100 ± 0
CNP 2 (n = 10), ρ(Cu) = 3.75 μg/ml	1.39 ± 0.19*	4.33 ± 0.16*	30.81 ± 3.45*	68.24 ± 5.53	100 ± 0
CNP 3 (n = 10), ρ(Cu) = 7.50 μg/ml	12.02 ± 0.95*	16.94 ± 1.85*	29.62 ± 3.22*	69.19 ± 7.23	100 ± 0
CNP 4 (n = 10), ρ(Cu) = 12.32 μg/ml	11.19 ± 1.55*	19.78 ± 1.43*	48.77 ± 5.05*	89.18 ± 7.85*	100 ± 0

Note: * $P \le 0.05$ as compared with the reference; CNP: copper nanoparticles.

lla typhimurium 144 starting with concentration 33.46 μg/ml. The complete inhibition of growth of *Bacillus subtilis ATCC* 6633 has been reported at higher concentrations, starting with 133.8 μg/ ml, which is an important result, since it is known that *Bacillus subtilis* bacteria are components of normal human microflora [13]. It is known that the *Bacillus* bacteria may have resistance to silver nitrate, therefore, they are used for the biological synthesis of silver nanoparticles. The mechanism of *Bacillus* resistance to silver has not been studied completely and is associated with the presence of nitrate reductase enzyme in the bacteria [3].

The prominent antimicrobial effect of silver nanoparticles has been established in relation to antibiotic-resistant clinical isolates, as complete inhibition of growth has been reported for all strains. The results have showed that clinical isolates, such as *P. aeruginosa* and *K. ozaenae,* characterized by resistance to the majority of antibiotics are sensitive to silver nanoparticles. The diameters of retardation zones for *P. aeruginosa* are the highest among all strains studied. Both the gram-negative *Enterobacteriacae* bacteria (*K. ozaenae, E. aerogenes, C. freundii E.coli*, and *P. mirabilis*) and the gram-positive *S. aureus* coccal bacteria appear to be sensitive to silver nanoparticles. The effective concentration of silver nanoparticles is 0.03–0.13 μg/ mm^2 per unit of the culture medium surface area. No secondary growth has been reported in all areas of retardation after 15 days of observation.

The mechanism of antimicrobial action of silver nanoparticles has not been studied enough. There is a widespread view that the impact of nanoparticles is associated with the formation of active oxygen that causes oxidative stress within the cell [14].

The *fine silica—silver clusters* nanocomposite (FSSC) (12–18 nm) obtained by the mechanic sorption technique has high antimicrobial effect on *S. aureus, E. coli, C. albicans*, and *P. aeruginosa*. The highest antimicrobial activity has been reported for *E. coli*., with minimum inhibitory concentration (MIC) being 330 μg/ml and minimum bactericidal concentration being 660 μg/ml. The experiment has showed that resistance of the gram-negative and the gram-positive bacteria to FSSC silver nanoparticles depends on the structure of microbial cells, physical and chemical processes occurring in the course of interaction of nanoparticles with cells, and on other factors to be studied further.

The FSSC sorption properties, especially with respect to the low- and medium-molecular compounds have been found to be better as compared with activated charcoal, Sorbex, and Silix: for initial concentration of methylene blue 10 mg/ ml, all the above sorbents bind it with the following rates: activated charcoal 130 mg/g; Sorbex 110 mg/g; Silix 45 mg/g; and FSSC 280 mg/g.

The development of drug dosage forms is very important during the preclinical studies of potential drugs. The Danylo of Galicia Lviv National Medical University, the P.L. Shupyk National Medical Academy of Postgraduate Education, and the O.O. Bogomolets National Medical University have developed drug dosage forms for administering silver and copper nanoparticles orally and intravenously.

Thus, the nanoparticles of silver, copper, and iron have antimicrobial effect that makes it possible to use them for the treatment of infectious diseases. The acute toxicity of metal nanoparticles depends on their nature, way of administration, and sex of laboratory animals. The nanoparticle effect on the heart activity, the hemodynamic status, and the osmotic resistance of erythrocyte membranes depend on the dose administered.

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ФАРМАКОЛОГІЧНІ ВЛАСТИВОСТІ НАНОМЕТАЛІВ: СРІБЛА, МІДІ, ЗАЛІЗА

В оглядовій статті узагальнені результати проведених досліджень з вивчення фармакологічних, токсикологічних та специфічних властивостей нанометалів (срібла, заліза, міді). Наночастинки срібла, міді, заліза проявляють протимікробну дію. Гостра токсичність нанометалів залежить від їх природи, шляху введення, а також статі тварин. Вплив на діяльність серця та стан гемодинаміки, а також на осмотичну резистентність мембран еритроцитів має дозозалежний характер.

Ключові слова: наночастинки, наносрібло, наномідь, нанозалізо, властивості.

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В обзорной статье обобщены результаты проведенных исследований по изучению фармакологических, токсикологических и специфических свойств нанометаллов (серебра, железа, меди). Наночастицы серебра, меди, железа проявляют противомикробное действие. Острая токсичность нанометаллов зависит от их природы, пути введения, а также пола животных. Влияние на деятельность сердца и состояние гемодинамики, а также на осмотическую резистентность мембран эритроцитов имеет дозозависимый характер.

Ключевые слова: наночастицы, наносеребро, наномедь, наножелезо, свойства.

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